1262151

THE UNITED STATES OF AMERICA

TO ALL TO WHOM THESE PRESENTS SHALL COME:

UNITED STATES DEPARTMENT OF COMMERCE

United States Patent and Trademark Office

December 15, 2004

THIS IS TO CERTIFY THAT ANNEXED HERETO IS A TRUE COPY FROM THE RECORDS OF THE UNITED STATES PATENT AND TRADEMARK OFFICE OF THOSE PAPERS OF THE BELOW IDENTIFIED PATENT APPLICATION THAT MET THE REQUIREMENTS TO BE GRANTED A FILING DATE.

APPLICATION NUMBER: *60/517,243*
FILING DATE: *November 04, 2003*
RELATED PCT APPLICATION NUMBER: *PCT/US04/36663*

Certified by

Jon W Dudas

Acting Under Secretary of Commerce
for Intellectual Property
and Acting Director of the U.S.
Patent and Trademark Office

16638 U.S.PTO

"EXPRESS MAIL CERTIFICATE"

"Express Mail" Mailing Label Number EL964433998US

Date Of Deposit: November 4, 2003

I Hereby Certify That This Paper Or Fee Is Being Deposited With The United States Postal Service "Express Mail Post Office To Addressee" Service Under 37 CFR 1.10 On The Date Indicated Above And Is Addressed To: COMMISSIONER FOR PATENTS, MAIL STOP: PROVISIONAL APPLICATION, P.O. BOX 1450, ALEXANDRIA, VA 22313-1450

Name Of Person Mailing Paper Or Fee

(Type Or Print) Shawn Simpson

Signature

03917 U.S.PTO 60/517243 110403

PROVISIONAL APPLICATION COVER SHEET

This is a request for filing a PROVISIONAL APPLICATION for PATENT under 37 CFR 1.53(c).

Docket No.	PU60574P		

INVENTOR(s) / APPLICANT(s)

Last Name	First Name	Middle Initial	Residence (City and Either State or Foreign Country)
WAN	Zehong	.	King of Prussia, Pennsylvania
YAN	Hongxing	.	King of Prussia, Pennsylvania
PALOVICH	Michael	R.	King of Prussia, Pennsylvania

TITLE OF THE INVENTION (280 characters max)

M3 MUSCARINIC ACETYLCHOLINE RECEPTOR ANTAGONISTS

Correspondence Address:

GLAXOSMITHKLINE
Corporate Intellectual Property - UW2220
709 Swedeland Road
King of Prussia

Telephone No. 610-270-5019
Facsimile No. 610-270-5090

State	PA	Zip Code	19406-0939	Country	United States of America

ENCLOSED APPLICATION PARTS (check all that apply)

☒ Specification	Number of Pages	23	**Total Number of Pages = 24**
☒ Abstract	Number of Pages	1	
☐ Drawings	Number of Sheets		☐ Other (specify)

METHOD OF PAYMENT OF FILING FEES FOR THIS PROVISIONAL APPLICATION FOR PATENT

☒ The Commissioner is hereby authorized to charge filing fees and credit Deposit Account No. **19-2570**	PROVISIONAL FILING FEE AMOUNT ($)	**$160.00**

Respectfully submitted,
Signature:

Soma G. Simon

Date:

Registration No.: 37,444

☐ Additional inventors are being named on separately numbered sheets attached hereto.

PROVISIONAL APPLICATION FILING ONLY

SEND TO: Commissioner for Patents, P.O. Box 1450, Mail Stop: Patent Application, Alexandria, VA 22313-1450.

20462

CUSTOMER NUMBER

N:\SOMA\CASES\PU60574P\ProvApp.doc

M_3 MUSCARINIC ACETYLCHOLINE RECEPTOR ANTAGONISTS

FIELD OF THE INVENTION

This invention relates to novel derivatives of 8-azoniabicyclo [3,2,1]octanes, pharmaceutical compositions, processes for their preparation, and use thereof in treating M_3 muscarinic acetylcholine receptor mediated diseases.

BACKGROUND OF THE INVENTION

Acetylcholine released from cholinergic neurons in the peripheral and central nervous systems affects many different biological processes through interaction with two major classes of acetylcholine receptors – the nicotinic and the muscarinic acetylcholine receptors. Muscarinic acetylcholine receptors (mAChRs) belong to the superfamily of G-protein coupled receptors that have seven transmembrane domains. There are five subtypes of mAChRs, termed M_1-M_5, and each is the product of a distinct gene. Each of these five subtypes displays unique pharmacological properties. Muscarinic acetylcholine receptors are widely distributed in vertebrate organs, and these receptors can mediate both inhibitory and excitatory actions. For example, in smooth muscle found in the airways, bladder and gastrointestinal tract, M_3 mAChRs mediate contractile responses. For review, please see {Brown 1989 247 /id}.

Muscarinic acetylcholine receptor dysfunction has been noted in a variety of different pathophysiological states. For instance, in asthma and chronic obstructive pulmonary disease (COPD), inflammatory conditions lead to loss of inhibitory M_2 muscarinic acetylcholine autoreceptor function on parasympathetic nerves supplying the pulmonary smooth muscle, causing increased acetylcholine release following vagal nerve stimulation. This mAChR dysfunction results in airway hyperreactivity mediated by increased stimulation of M_3 mAChRs{Costello, Evans, et al. 1999 72 /id}{Minette, Lammers, et al. 1989 248 /id}. Similarly, inflammation of the gastrointestinal tract in inflammatory bowel disease (IBD) results in M_3

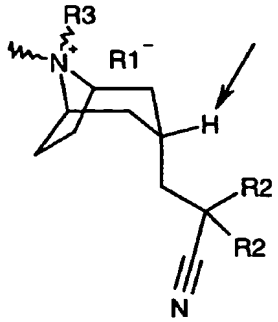
mAChR-mediated hypermotility {Oprins, Meijer, et al. 2000 245 /id}. Incontinence due to bladder hypercontractility has also been demonstrated to be mediated through increased stimulation of M_3 mAChRs {Hegde & Eglen 1999 251 /id}. Thus the identification of subtytpe-selective mAChR antagonists may be useful as therapeutics in these mAChR-mediated diseases.

Despite the large body of evidence supporting the use of anti-muscarinic receptor therapy for treatment of a variety of disease states, relatively few anti-muscarinic compounds are in use in the clinic. Thus, there remains a need for novel compounds that are capable of causing blockade at M_3 mAChRs. Conditions associated with an increase in stimulation of M_3 mAChRs, such as asthma, COPD, IBD and urinary incontinence would benefit by compounds that are inhibitors of mAChR binding.

SUMMARY OF THE INVENTION

This invention provides for a method of treating a muscarinic acetylcholine receptor (mAChR) mediated disease, wherein acetylcholine binds to an M_3 mAChR and which method comprises administering an effective amount of a compound of Formula (I) or a pharmaceutically acceptable salt thereof.

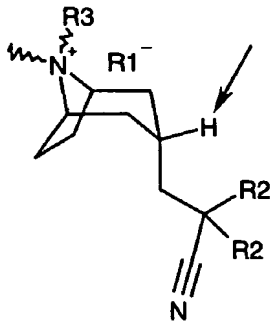
This invention also relates to a method of inhibiting the binding of acetylcholine to its receptors in a mammal in need thereof which comprises administering to aforementioned mammal an effective amount of a compound of Formula (I).



(I)

DETAILED DESCRIPTION OF THE INVENTION

This invention relates to novel derivatives of 8-azoniabicyclo [3,2,1]octanes, pharmaceutical compositions, processes for their preparation, and use thereof in treating M_3 muscarinic acetylcholine receptor mediated diseases.



(I)

Compounds with Formula (I) are preferred, wherein:

The H atom indicated is in the exo position;

$R1^-$ represents an anion associated with the positive charge of the N atom. $R1^-$ may be but is not limited to chloride, bromide, iodide, sulfate, benzene sulfonate and toluene sulfonate;

R2 is selected from the group consisting of straight or branched chain lower alkyl groups (having preferably from 1 to 6 carbon atoms), cycloalkyl groups (having from 5 to 6 carbon atoms), cycloalkyl-alkyl (having 6 to 10 carbon atoms), heterocycloalkyl (having 5 to 6 carbon atoms) and N or O as the heteroatom, heterocycloalkyl-alkyl (having 6 to10 carbon atoms) and N or O as the heteroatom, aryl, optionally substituted aryl, heteroaryl, and optionally substituted heteroaryl;

R3 is selected from the group consisting of (C_2-C_{12})alkyl, (C_1-C_6)alkenyl, (C_1-C_6)alkyl(C_3-C_6)cycloalkyl, (C_1-C_6)alkyl-phenyl, (C_1-C_6)alkyl-OH, (C_1-C_6)alkyl-CN, (C_1-C_6)alkyl-halogen, (C_1-C_6)alkyl-CF_3, (C_1-C_6)alkyl-OCH_3, (C_1-C_6)alkyl-O-(C_1-C_6)alkyl-OCH_3.

All of the aryl, heteroaryl, and heterocyclic containing moieties may be optionally substituted as defined herein below.

For use herein the term "the aryl, heteroaryl, and heterocyclic containing moieties" refers to both the ring and the alkyl, or if included, the alkenyl rings, such as aryl, arylalkyl, and aryl alkenyl rings. The term "moieties" and "rings" may be interchangeably used throughout.

As used herein, "optionally substituted" unless specifically defined shall mean such groups as halogen, such as fluorine, chlorine, bromine or iodine; hydroxy; hydroxy substituted C_{1-10}alkyl; C_{1-10} alkoxy, such as methoxy or ethoxy; $S(O)_{m'}$ C_{1-10} alkyl, wherein m' is 0, 1 or 2, such as methyl thio, methyl sulfinyl or methyl sulfonyl; amino, mono & di-substituted amino, such as in the NR_4R_5 group; $NHC(O)R_4$; $C(O)NR_4R_5$; C(O)OH; $S(O)_2NR_4R_5$; $NHS(O)_2R_4$, C_{1-10} alkyl, such as methyl, ethyl, propyl, isopropyl, or t-butyl; halosubstituted C_{1-10} alkyl, such as CF_3; an optionally substituted aryl, such as phenyl, or an optionally substituted arylalkyl, such as benzyl or phenethyl, optionally substituted heterocylic, optionally substituted heterocyclicalkyl, optionally substituted heteroaryl, optionally substituted heteroaryl alkyl, wherein these aryl , heteroaryl, or heterocyclic moieties may be substituted one to two times by halogen; hydroxy; hydroxy substituted alkyl; C_{1-10} alkoxy; $S(O)_{m'}C_{1-10}$ alkyl; amino, mono & di-substituted alkyl amino, such as in the NR_4R_5 group; C_{1-10} alkyl, or halosubstituted C_{1-10} alkyl, such as CF_3.

Suitable pharmaceutically acceptable salts are well known to those skilled in the art and include basic salts of inorganic and organic acids, such as hydrochloric acid, hydrobromic acid, sulphuric acid, phosphoric acid, methane sulphonic acid, ethane sulphonic acid, acetic acid, malic acid, tartaric acid, citric acid, lactic acid, oxalic acid, succinic acid, fumaric acid,

maleic acid, benzoic acid, salicylic acid, phenylacetic acid and mandelic acid. In addition, pharmaceutically acceptable salts of compounds of Formula (I) may also be formed with a pharmaceutically acceptable cation. Suitable pharmaceutically acceptable cations are well known to those skilled in the art and include alkaline, alkaline earth, ammonium and quaternary ammonium cations.

The following terms, as used herein, refer to:

- "halo" - all halogens, that is chloro, fluoro, bromo and iodo.
- "C_{1-10}alkyl" or "alkyl" - both straight and branched chain moieties of 1 to 10 carbon atoms, unless the chain length is otherwise limited, including, but not limited to, methyl, ethyl, *n*-propyl, *iso*-propyl, *n*-butyl, *sec*-butyl, *iso*-butyl, *tert*-butyl, *n*-pentyl and the like.
- "cycloalkyl" is used herein to mean cyclic moiety, preferably of 3 to 8 carbons, including but not limited to cyclopropyl, cyclopentyl, cyclohexyl, and the like.
- "alkenyl" is used herein at all occurrences to mean straight or branched chain moiety of 2-10 carbon atoms, unless the chain length is limited thereto, including, but not limited to ethenyl, 1-propenyl, 2-propenyl, 2-methyl-1-propenyl, 1-butenyl, 2-butenyl and the like.
- "aryl" - phenyl and naphthyl;
- "heteroaryl" (on its own or in any combination, such as "heteroaryloxy", or "heteroaryl alkyl") - a 5-10 membered aromatic ring system in which one or more rings contain one or more heteroatoms selected from the group consisting of N, O or S, such as, but not limited, to pyrrole, pyrazole, furan, thiophene, quinoline, isoquinoline, quinazolinyl, pyridine, pyrimidine, oxazole, tetrazole, thiazole, thiadiazole, triazole, imidazole, or benzimidazole.
- "heterocyclic" (on its own or in any combination, such as "heterocyclicalkyl" or "heterocycloalkyl") - a saturated or partially unsaturated 4-10 membered ring system in which one or more rings contain one or more heteroatoms selected from the group consisting of N, O, or S; such as, but not limited to, pyrrolidine, piperidine, piperazine, morpholine,

tetrahydropyran, thiomorpholine, or imidazolidine. Furthermore, sulfur may be optionally oxidized to the sulfone or the sulfoxide.

• "arylalkyl" or "heteroarylalkyl" or "heterocyclicalkyl" is used herein to mean C_{1-10} alkyl, as defined above, attached to an aryl, heteroaryl or heterocyclic moiety, as also defined herein, unless otherwise indicated.

• "sulfinyl" - the oxide S (O) of the corresponding sulfide, the term "thio" refers to the sulfide, and the term "sulfonyl" refers to the fully oxidized $S(O)_2$ moiety.

• Preferred compounds useful in the present invention include:
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(cyclohexylmethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(cyclopropylmethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-8-butyl-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-8-(4-chlorobutyl)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-dodecyl-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(2-propen-1-yl)-8-azoniabicyclo[3.2.1]octane iodide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(phenylmethyl)-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(2-hydroxyethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-ethyl-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-propyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(5-hexen-1-yl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;

(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(4,4,4-trifluorobutyl)-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(3-phenylpropyl)-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(2-cyclohexylethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(3-cyanopropyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-[2-(methyloxy)ethyl]-8-azoniabicyclo[3.2.1]octane bromide; and
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(2-{[2-(methyloxy)ethyl]oxy}ethyl)-8-azoniabicyclo[3.2.1]octane bromide.

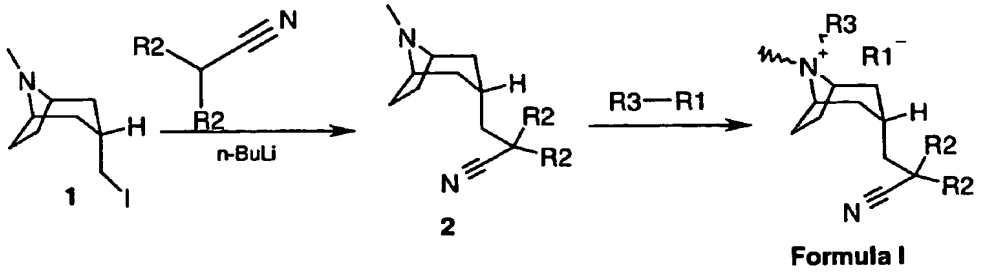
Methods of Preparation

Preparation

The compounds of Formula (I) may be obtained by applying synthetic procedures, some of which are illustrated in the Schemes below. The synthesis provided for these Schemes is applicable for producing compounds of Formula (I) having a variety of different R1, R2 and R3 which are reacted, employing substituents which are suitable protected, to achieve compatibility with the reactions outlined herein. Subsequent deprotection, in those cases, then affords compounds of the nature generally disclosed. While some Schemes are shown with specific compounds, this is merely for illustration purpose only.

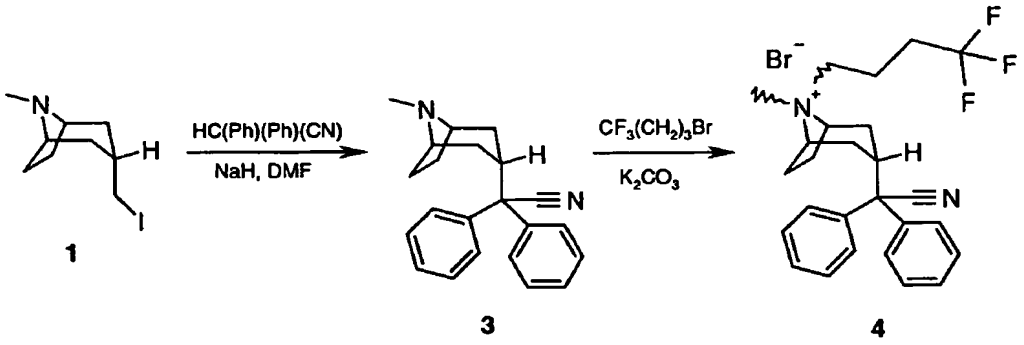
The general preparation method is shown in **Scheme I.** The synthesis started with compound **1**. Coupling reaction with the anion derived from HC(CN)(R2)(R2) provided **2**. Treatment with R3-R1 then furnished the quaternary ammonium salt with Formula (I).

Scheme I.



A more spercific preparation method leading to compounds with Formula (I) is outlined in **Scheme II**. Alkylation of diphenylacetonitrile with **1** afforded compound **3**. Treatment with $CF_3(CH_2)_3Br$ then afforded quarternary ammonium salt **4.**

Scheme II



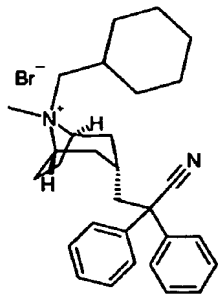
SYNTHETIC EXAMPLES

The following examples are provided as illustrative of the present invention but not limiting in any way:

General Preparation Procedures

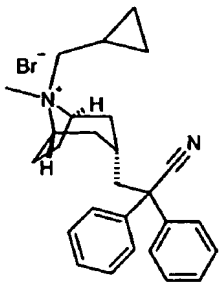
A solution of 3-((3-endo)-8-methyl-8-aza-bicyclo[3.2.1]oct-3-yl)-2,2-diphenyl-propionitrile (33.0 mg, 0.10 mmol) in CH_2Cl_2 (0.5 mL) and MeCN (0.5 mL) was mixed with RBr (1.0 mmol) and K_2CO_3 (27.6 mg, 0.20 mmol). The resultant mixture was stirred at room temperature for certain reaction time (specified in following examples). It was then diluted with DMSO (0.3 mL) and concentrated. Purification via a reverse phase HPLC (Gilson) afforded the target compound.

Example 1



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(cyclohexylmethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromideThe title compound was prepared in 15% yield by following the general experimental procedure (reaction time = 7 days): LCMS (ES) *m/z* 427 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.29 (m, 3H), 1.43 (m, 2H), 1.83 (m, 8H), 2.19 (m, 1H), 2.42 (m, 6H), 3.00 (m, 2H), 3.04 (s, 3H), 3.10 (d, 2H), 3.84 (s, 2H), 7.35 (m, 2H), 7.43 (m, 4H), 7.49 (m, 4H).

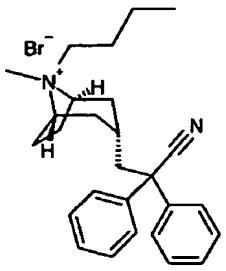
Example 2



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(cyclopropylmethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 51% yield by following the general experimental procedure (reaction time = 70 hours): LCMS (ES) *m/z* 385 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 0.48 (m, 2H), 0.83 (m, 2H), 1.13 (m, 1H), 1.82 (m, 2H), 2.22 (m, 1H), 2.42 (m, 6H), 3.01 (m, 2H), 3.12 (m, 3H), 3.19 (d, 2H), 3.90 (m, 2H), 7.35 (m, 2H), 7.43 (m, 4H), 7.50 (m, 4H).

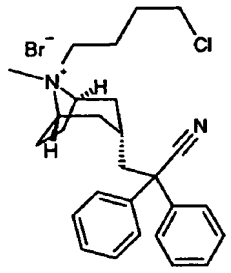
Example 3



(3-endo)-8-butyl-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 26% yield by following the general experimental procedure (reaction time = 70 hours): LCMS (ES) *m/z* 387 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.04 (m, 3H), 1.45 (m, 2H), 1.74 (m, 2H), 1.84 (d, 2H), 2.21 (m, 1H), 2.45 (m, 6H), 3.00 (m, 2H), 3.02 (s, 3H), 3.20 (m, 2H), 3.83 (s, 2H), 7.35 (m, 2H), 7.42 (m, 4H), 7.49 (m, 4H).

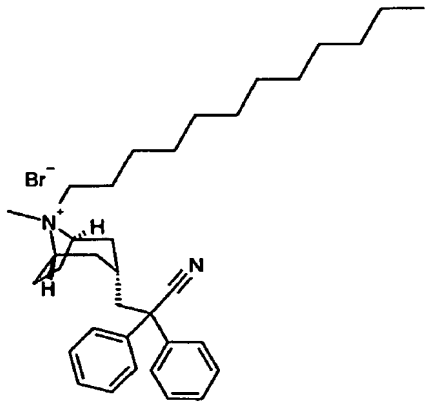
Exampl 4



(3-endo)-8-(4-chlorobutyl)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 37% yield by following the general experimental procedure (reaction time = 70 hours): LCMS (ES) *m/z* 421 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.88 (m, 6H), 2.18 (m, 1H), 2.45 (m, 6H), 3.01 (m, 2H), 3.04 (s, 3H), 3.28 (m, 2H), 3.67 (m, 2H), 3.84 (s, 2H), 7.35 (m, 2H), 7.42 (m, 4H), 7.49 (m, 4H).

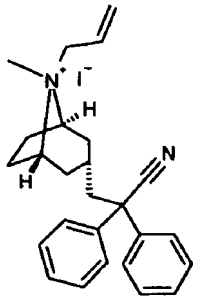
Example 5



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-dodecyl-8-methyl-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 10% yield by following the general experimental procedure (reaction time = 70 hours): LCMS (ES) *m/z* 499 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 0.95 (t, 3H), 1.36 (m, 18H), 1.73 (m, 2H), 1.82 (d, 2H), 2.18 (m, 1H), 2.46 (m, 6H), 3.00 (d, 2H), 3.02 (s, 3H), 3.19 (m, 2H), 3.82 (s, 2H), 7.35 (m, 2H), 7.42 (m, 4H), 7.49 (m, 4H).

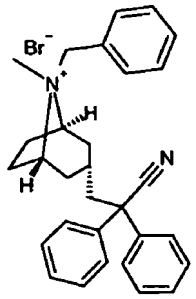
Exampl 6



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(2-propen-1-yl)-8-azoniabicyclo[3.2.1]octane iodide

The title compound was prepared in 37% yield by following the general experimental procedure (reaction time = 3 hours): LCMS (ES) *m/z* 371 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.83 (m, 2H), 2.20 (m, 1H), 2.36 (m, 2H), 2.47 (m, 4H), 3.02 (m, 5H), 3.85 (s, 2H), 3.92 (d, 2H), 5.71 (m, 2H), 6.09 (m, 1H), 7.35 (m, 2H), 7.42 (m, 4H), 7.51 (m, 4H).

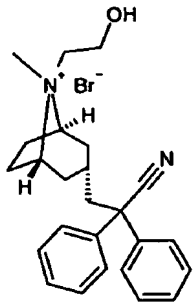
Example 7



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(phenylmethyl)-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 39% yield by following the general experimental procedure (reaction time = 3 hours): LCMS (ES) *m/z* 421 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.85 (d, 2H), 2.17 (m, 1H), 2.45 (m, 4H), 2.73 (m, 2H), 2.92 (s, 3H), 3.04 (d, 2H), 3.86 (s, 2H), 4.45 (s, 2H), 7.34 (m, 2H), 7.42 (m, 4H), 7.48 (m, 4H), 7.56 (m, 5H).

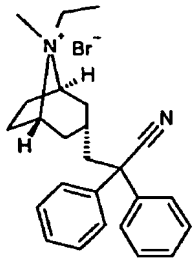
Example 8



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(2-hydroxyethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 40% yield by following the general experimental procedure (reaction time = 10 days): LCMS (ES) *m/z* 375 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.84 (m, 2H), 2.04 (m, 1H), 2.22 (m, 2H), 2.34 (m, 2H), 2.50 (m, 2H), 2.74 (s, 1H), 2.95 (d, 1H), 3.01 (d, 1H), 3.14 (s, 1H), 3.33 (s, 3H), 3.40 (m, 1H), 3.81 (m, 1H), 3.99 (m, 2H), 7.35 (m, 2H), 7.42 (m, 4H), 7.48 (m, 4H).

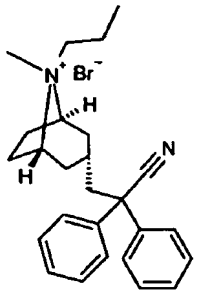
Example 9



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-ethyl-8-methyl-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 60% yield by following the general experimental procedure (reaction time = 70 hours): LCMS (ES) *m/z* 359 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.32 (t, 3H), 1.83 (d, 2H), 2.21 (m, 1H), 2.36 (m, 4H), 2.52 (m, 2H), 3.02 (m, 5H), 3.35 (m, 2H), 3.81 (s, 2H), 7.35 (m, 2H), 7.42 (m, 4H), 7.49 (m, 4H).

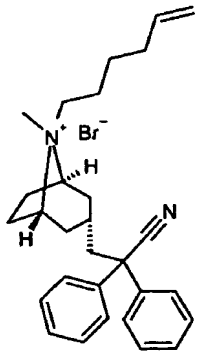
Example 10



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-propyl-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 20% yield by following the general experimental procedure (reaction time = 70 hours): LCMS (ES) *m/z* 373 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.02 (t, 3H), 1.74 (m, 1H), 1.81 (d, 2H), 2.18 (m, 1H), 2.34 (m, 2H), 2.45 (m, 4H), 3.00 (m, 2H), 3.02 (s, 3H), 3.17 (m, 2H), 3.82 (s, 2H), 7.35 (m, 2H), 7.42 (m, 4H), 7.49 (m, 4H).

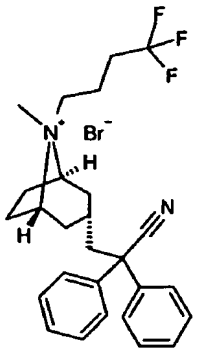
Example 11



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(5-hexen-1-yl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 29% yield by following the general experimental procedure (reaction time = 70 hours): LCMS (ES) *m/z* 413 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.48 (m, 2H), 1.79 (m, 4H), 2.18 (m, 3H), 2.42 (m, 6H), 3.00 (m, 5H), 3.22 (m, 2H), 3.82 (s, 2H), 5.05 (m, 2H), 5.86 (m, 1H), 7.35 (m, 2H), 7.42 (m, 4H), 7.49 (m, 4H).

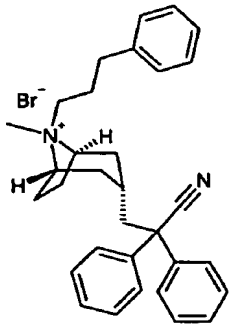
Exampl 12



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(4,4,4-trifluorobutyl)-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 23% yield by following the general experimental procedure (reaction time = 70 hours): LCMS (ES) *m/z* 441 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.80 (d, 2H), 2.03 (m, 2H), 2.19 (m, 1H), 2.41 (m, 8H), 3.01 (m, 2H), 3.06 (s, 3H), 3.30 (m, 2H), 3.87 (s, 2H), 7.35 (m, 2H), 7.42 (m, 4H), 7.49 (m, 4H).

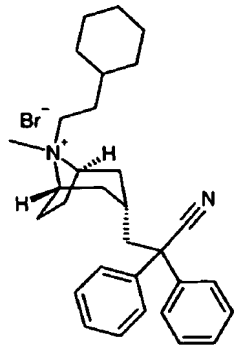
Example 13



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(3-phenylpropyl)-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 43% yield by following the general experimental procedure (reaction time = 7 days): LCMS (ES) *m/z* 449 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.78 (d, 2H), 2.07 (m, 2H), 2.26 (m, 4H), 2.46 (m, 3H), 2.71 (t, 2H), 2.97 (d, 2H), 2.99 (s, 3H), 3.21 (m, 2H), 3.80 (s, 2H), 7.25 (m, 2H), 7.31 (m, 4H), 7.41 (m, 5H), 7.47 (m, 4H).

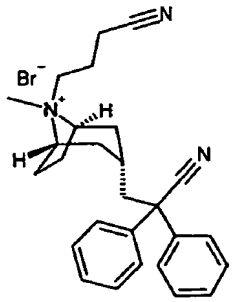
Example 14



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(2-cyclohexylethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 21% yield by following the general experimental procedure (reaction time = 7 days): LCMS (ES) *m/z* 441 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.04 (m, 2H); 1.29 (m, 4H), 1.62 (m, 2H), 1.75 (m, 7H), 2.18 (m, 1H), 2.34 (m, 4H), 2.49 (m, 2H), 3.00 (m, 5H), 3.24 (m, 2H), 3.82 (s, 2H), 7.35 (m, 2H), 7.42 (m, 4H), 7.49 (m, 4H).

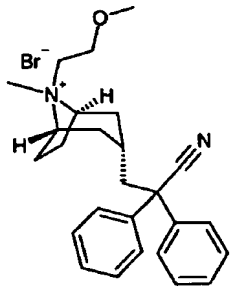
Example 15



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(3-cyanopropyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 48% yield by following the general experimental procedure (reaction time = 7 days): LCMS (ES) *m/z* 398 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.83 (d, 2H), 2.17 (m, 3H), 2.42 (m, 6H), 2.60 (t, 2H), 3.01 (m, 2H), 3.06 (s, 3H), 3.32 (m, 2H), 3.87 (s, 2H), 7.35 (m, 2H), 7.42 (m, 4H), 7.49 (m, 4H).

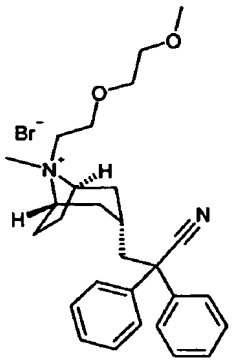
Example 16



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-[2-(methyloxy)ethyl]-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 24% yield by following the general experimental procedure (reaction time = 7 days): LCMS (ES) *m/z* 389 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.80 (d, 2H), 2.18 (m, 1H), 2.34 (m, 2H), 2.48 (m, 4H), 3.00 (m, 2H), 3.10 (s, 3H), 3.38 (s, 3H), 3.50 (m, 2H), 3.81 (m, 2H), 3.93 (s, 2H), 7.35 (m, 2H), 7.42 (m, 4H), 7.49 (m, 4H).

Example 17



(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(2-{[2-(methyloxy)ethyl]oxy}ethyl)-8-azoniabicyclo[3.2.1]octane bromide

The title compound was prepared in 28% yield by following the general experimental procedure (reaction time = 7 days): LCMS (ES) *m/z* 433 $(M)^+$; ^{1}H-NMR($CDCl_3$) δ 1.80 (d, 2H), 2.18 (m, 1H), 2.34 (m, 2H), 2.48 (m, 4H), 3.01 (m, 2H), 3.11 (s, 3H), 3.36 (s, 3H), 3.50 (m, 2H), 3.56 (m, 2H), 3.64 (m, 2H), 3.91 (m, 2H), 3.96 (s, 2H), 7.35 (m, 2H), 7.43 (m, 4H), 7.49 (m, 4H).

BIOLOGICAL EXAMPLES

The inhibitory effects of compounds at the M_3 mAChR of the present invention are determined by the following *in vitro* and *in vivo* assay:

Analysis of Inhibition of Receptor Activation by Calcium Mobilization: Stimulation of mAChRs expressed on CHO cells were analyzed by monitoring receptor-activated calcium mobilization as previously described[10]. CHO cells stably expressing M_3 mAChRs were plated in 96 well black wall/clear bottom plates. After 18 to 24 hours, media was aspirated and replaced with 100 µl of load media (EMEM with Earl's salts, 0.1% RIA-grade BSA (Sigma, St. Louis MO), and 4 µM Fluo-3-acetoxymethyl ester fluorescent indicator dye (Fluo-3 AM, Molecular Probes, Eugene, OR) and incubated 1 hr at 37° C. The dye-containing media was then aspirated, replaced with fresh media (without Fluo-3 AM), and cells were incubated for 10 minutes at 37° C. Cells were then washed 3 times and incubated for 10 minutes at 37° C in 100 µl of assay buffer (0.1% gelatin (Sigma), 120 mM NaCl, 4.6 mM KCl, 1 mM KH_2PO_4, 25 mM $NaHCO_3$, 1.0 mM $CaCl_2$, 1.1 mM $MgCl_2$, 11 mM glucose, 20mM HEPES (pH 7.4)). 50 µl of compound ($1x10^{-11}$ – $1x10^{-5}$ M final in the assay) was added and the plates were incubated for 10 min. at 37° C. Plates were then placed into a fluorescent light intensity plate reader (FLIPR, Molecular Probes) where the dye loaded cells were exposed to excitation light (488 nm) from a 6 watt argon laser. Cells were activated by adding 50 µl of acetylcholine (0.1-10 nM final), prepared in buffer containing 0.1% BSA, at a rate of 50 µl/sec. Calcium mobilization, monitored as change in cytosolic calcium concentration, was measured as change in 566 nm emission intensity. The change in emission intensity is directly related to cytosolic calcium levels [11]. The emitted fluorescence from all 96 wells is measured simultaneously using a cooled

CCD camera. Data points are collected every second. This data was then plotting and analyzed using GraphPad PRISM software.

Methacholine-induced bronchoconstriction

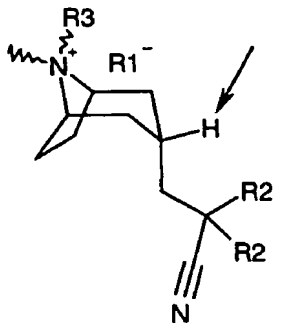
Airway responsiveness to methacholine was determined in awake, unrestrained BalbC mice (n = 6 each group). Barometric plethysmography was used to measure enhanced pause (Penh), a unitless measure that has been shown to correlate with the changes in airway resistance that occur during bronchial challenge with methacholine[12]. Mice were pretreated with 50 µl of compound (0.003-10 µg/mouse) in 50 µl of vehicle (10% DMSO) intranasally, and were then placed in the plethysmography chamber. Once in the chamber, the mice were allowed to equilibrate for 10 min before taking a baseline Penh measurement for 5 minutes. Mice were then challenged with an aerosol of methacholine (10 mg/ml) for 2 minutes. Penh was recorded continuously for 7 min starting at the inception of the methacholine aerosol, and continuing for 5 minutes afterward. Data for each mouse were analyzed and plotted by using GraphPad PRISM software.

All publications, including but not limited to patents and patent applications, cited in this specification are herein incorporated by reference as if each individual publication were specifically and individually indicated to be incorporated by reference herein as though fully set forth.

The above description fully discloses the invention including preferred embodiments thereof. Modifications and improvements of the embodiments specifically disclosed herein are within the scope of the following claims. Without further elaboration, it is believed that one skilled in the art can, using the preceding description, utilize the present invention to its fullest extent. Therefore the Examples herein are to be construed as merely illustrative and not a limitation of the scope of the present invention in any way. The embodiments of the invention in which an exclusive property or privilege is claimed are defined as follows.

What is claimed is:

1. A compound having structure I as indicated below:



(I)

wherein:

$R1^-$ represents an anion associated with the positive charge of the N atom; and

R2 is selected from the group consisting of straight or branched chain lower alkyl groups (having preferably from 1 to 6 carbon atoms), cycloalkyl groups (having from 5 to 6 carbon atoms), cycloalkyl-alkyl (having 6 to 10 carbon atoms), heterocycloalkyl (having 5 to 6 carbon atoms) and N or O as the heteroatom, heterocycloalkyl-alkyl (having 6 to10 carbon atoms) and N or O as the heteroatom, aryl, optionally substituted aryl, heteroaryl, and optionally substituted heteroaryl;

R3 is selected from the group consisting of $(C_2\text{-}C_{12})$alkyl, $(C_1\text{-}C_6)$alkenyl, $(C_1\text{-}C_6)$alkyl$(C_3\text{-}C_6)$cycloalkyl, $(C_1\text{-}C_6)$alkyl-phenyl, $(C_1\text{-}C_6)$alkyl-OH, $(C_1\text{-}C_6)$alkyl-CN, $(C_1\text{-}C_6)$alkyl-halogen, $(C_1\text{-}C_6)$alkyl-CF_3, $(C_1\text{-}C_6)$alkyl-OCH_3, and $(C_1\text{-}C_6)$alkyl-O-$(C_1\text{-}C_6)$alkyl-OCH_3.

2. A compound according to claim 1 wherein the H atom indicated is in the exo position.

3. A compound according to claim 1 wherein $R1^-$ is selected from the group consisting of chloride, bromide, iodide, sulfate, benzene sulfonate and toluene sultonate.

4. A compound according claim 1 selected from the group consisting of:
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(cyclohexylmethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(cyclopropylmethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-8-butyl-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-8-(4-chlorobutyl)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-dodecyl-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(2-propen-1-yl)-8-azoniabicyclo[3.2.1]octane iodide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(phenylmethyl)-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(2-hydroxyethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-ethyl-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-propyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(5-hexen-1-yl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(4,4,4-trifluorobutyl)-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(3-phenylpropyl)-8-azoniabicyclo[3.2.1]octane bromide;

(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(2-cyclohexylethyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-(3-cyanopropyl)-8-methyl-8-azoniabicyclo[3.2.1]octane bromide;
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-[2-(methyloxy)ethyl]-8-azoniabicyclo[3.2.1]octane bromide; and
(3-endo)-3-(2-cyano-2,2-diphenylethyl)-8-methyl-8-(2-{[2-(methyloxy)ethyl]oxy}ethyl)-8-azoniabicyclo[3.2.1]octane bromide.

5. A pharmaceutical composition for the treatment of muscarinic acetylcholine receptor mediated diseases comprising a compound according to claim 1 and a pharmaceutically acceptable carrier thereof.

6. A method of inhibiting the binding of acetylcholine to its receptors in a mammal in need thereof comprising administering a safe and effective amount of a compound according to claim 1.

7. A method of treating a muscarinic acetylcholine receptor mediated disease, wherein acetylcholine binds to said receptor, comprising administering a safe and effective amount of a compound according to claim 1.

8. A method according to claim 7 wherein the disease is selected from the group consisting of chronic obstructive lung disease, chronic bronchitis, asthma, chronic respiratory obstruction, pulmonary fibrosis, pulmonary emphysema and allergic rhinitis.

9. A method according to claim 8 wherein administration is via inhalation via the mouth or nose.

10. A method according to claim 9 wherein administration is via a medicament dispenser selected from a reservoir dry powder inhaler, a multi-dose dry powder inhaler or a metered dose inhaler.

11. A method according to claim 10 wherein the compound is administered to a human and has a duration of action of 12 hours or more for a 1 mg dose.

12. A method according to claim 11 wherein the compound has a duration of action of 24 hours or more.

13. A method according to claim 12 wherein the compound has a duration of action of 36 hours or more.

ABSTRACT OF THE DISCLOSURE

Muscarinic Acetylcholine receptor antagonists and methods of using them are provided.

Document made available under the Patent Cooperation Treaty (PCT)

International application number: PCT/US04/036663

International filing date: 04 November 2004 (04.11.2004)

Document type: Certified copy of priority document

Document details: Country/Office: US
Number: 60/517,243
Filing date: 04 November 2003 (04.11.2003)

Date of receipt at the International Bureau: 23 December 2004 (23.12.2004)

Remark: Priority document submitted or transmitted to the International Bureau in compliance with Rule 17.1(a) or (b)

World Intellectual Property Organization (WIPO) - Geneva, Switzerland
Organisation Mondiale de la Propriété Intellectuelle (OMPI) - Genève, Suisse